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INVESTIGATIONS IN FISH CONTROL

29. Efficacy of Methylpentynol as an Anesthetic on Four Salmonids

30. Toxicity of Methylpentynol to Selected Fishes

31. Annotated Bibliography on Methylpentynol

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INVESTIGATIONS IN FISH CONTROL

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Continued on inside back cover--

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IN THIS COVER

	Pages
Efficacy of Methylpentynol as an Anesthetic on Four Salmonids, by Robert M. Howland and Richard A. Schoettger.	1-11
Toxicity of Methylpentynol to Selected Fishes, by Leif L. Marking	1-7
Annotated Bibliography on Methylpentynol, by Gerald E. Svendsen	1-7

INVESTIGATIONS IN FISH CONTROL

29. Efficacy of Methylpentynol as an Anesthetic on Four Salmonids

By Robert M. Howland and Richard A. Schoettger

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CONTENTS

	Page
Abstract	3
Methods and materials	4
Results	4
Efficacy	4
Effects of water quality	9
Repeated use of solutions	9
Effects of repeated anesthetization	9
Discussion	9
Conclusions	9
References	10

EFFICACY OF METHYLPENTYNOL AS AN ANESTHETIC ON FOUR SALMONIDS

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ABSTRACT.--Effective concentrations of methylpentynol for anesthetizing rainbow trout, brown trout, brook trout, and lake trout were determined by a series of tests. Concentrations of 1.5 to 8 parts per thousand induced anesthesia in 4 to 57 minutes. Increase in water temperature accelerated anesthesia. Changes in pH or in water hardness had no significant effect on the rate of anesthesia. Repeated anesthesia had little effect on rate of response. The efficacy of anesthetic solutions was reduced by continuous use. Approximately 1 kilogram of rainbow trout could be effectively narcotized per milliliter of drug. Methylpentynol was compared with MS-222 as a fish anesthetic. Fifty - times as much methylpentynol was necessary to yield the equivalent effect of 100 parts per million of MS-222. Methylpentynol may be more appropriate as a sedative or soporific for salmonids than as an anesthetic.

The demand for anesthetics in stripping, marking, and transporting fish has led to experimentation with various drugs used in human medicine, such as methylpentynol. Parkhurst and Smith (1957) found that the compound worked well to quiet fish for stripping. Since then, a number of investigators have found that methylpentynol is a potent fish anesthetic and is also useful as a sedative in fish transport (Norris et al., 1960; McFarland, 1959, 1960; Carlson, 1965). However, fish entering anesthesia may struggle violently and are not completely immobilized under narcosis, according to reports cited by Bell (1964). They may also go into respiratory arrest, thereby making the maintenance of anesthesia difficult (Klontz, 1964).

Methylpentynol (3-Methyl-1-pentyn-3-ol) was originally patented in 1913 by the Bayer Company of Germany. Some common synonyms of the compound include methylparafynol, Oblivon, and Dormison (Stecher et al., 1960). During the early 1950's it was introduced as a hypnotic in human medicine (Margolin et al., 1951; Hirsh and Orsinger, 1952; and Perlman and Johnson, 1952), but because of its side - effects and questionable efficacy (Lasagna, 1954; Marley, 1958; Kennedy and Marley, 1959; Goodman and Gilman, 1965) it had only a brief period of popularity.

Lennon (1967) discussed recent amendments to the Federal Food, Drug, and Cosmetic Act which require that chemicals used on fish be cleared and labeled for specific uses. We elected to investigate methylpentynol as one of the anesthetics because the treated fish may be eaten by humans. Information on the toxicity of methylpentynol to fish, its efficacy as a fish anesthetic, and its residues in fish tissues is necessary to obtain clearance.

We centered our investigation on the efficacy of methylpentynol, specifically the concentrations for practical use on rainbow trout, brown trout, brook trout, and lake trout. We also measured the effects of water quality, size of fish, repeated exposure, and reuse of anesthetic solutions on efficacy.

METHODS AND MATERIALS

We purchased a technical grade (Eastman grade) of methylpentynol from the Eastman Kodak Company, Rochester, N.Y. Its efficacy as a fish anesthetic was measured with trout of two size-ranges: 2 to 6 inches and 7 to 12 inches (table 1). The methods of preparing test solutions, waters of various qualities, controlling temperature, and acclimating the test fish were as described by Schoettger and Julin (1967).

The behavioral responses of trout to methylpentynol conformed reasonably well to those reported for fish exposed to MS-222 (Schoettger and Julin, 1967). Effectiveness of the drug was evaluated by placing the trout in various concentrations and recording the times for all of the test fish to exhibit the following behavioral responses: sedation; total loss of equilibrium, stage 11; and loss of reflex. We also recorded the time required for the first fish to enter medullary collapse. When all fish reached this stage, they were returned to well water for recovery.

Ten fish were used to bioassay each concentration, and in most instances the experiments were replicated. The amenability of trout to handling when in the deeper stages of narcosis was tested by removing them briefly from the solutions, and simulating the stripping procedure.

TABLE 1.--Species and sources of test fish

Species	Length (inches)	Source
Rainbow trout (*Salmo gairdneri*)	2-6	Raised at Fish Control Laboratory; eggs from Rainbow Ranches, Spokane, Washington
	7-12	NFH[1], Manchester, Iowa
Brown trout (*Salmo trutta*)	2-6	Raised at Fish Control Laboratory; eggs from McNenny NFH, Spearfish, South Dakota
	7-12	NFH, Manchester, Iowa
Brook trout (*Salvelinus fontinalis*)	2-6	Raised at Fish Control Laboratory; eggs from SFH[2], Osceola, Wisconsin
Lake trout (*Salvelinus namaycush*)	2-6	NFH, Jordan River, Elmira, Michigan

[1] National Fish Hatchery
[2] State Fish Hatchery

The reusability of methylpentynol solutions was determined by anesthetizing fresh groups of five rainbow trout, 7 to 12 inches long, in 5 liters of a 5 p.p.t. (part per thousand) solution at 12° C. This concentration anesthetizes rainbow trout to loss of reflex within 2 minutes. Fresh groups of fish were exposed until the drug was no longer effective within 2 minutes. We then calculated the kilograms of trout anesthetized per milliliter of the drug.

The effect of repeated exposure on the susceptibility of rainbow trout to methylpentynol was measured by anesthetizing 10 individuals daily for 10 consecutive days at 12° C. They were narcotized to loss of reflex in a concentration of 5 p.p.t. and then placed in flowing well water for recovery. Each day, the responses of single-exposure and untreated control fish were observed in conjunction with the group undergoing repeated exposure. After 10 days, we made a gross examination for pathology in the repeatedly exposed fish, a group treated only once, and an untreated group.

RESULTS

Efficacy

Methylpentynol was 100 percent effective at concentrations of 1.5 to 8 p.p.t. for anesthetizing rainbow trout, brown trout, brook trout, and lake trout (tables 2, 3, 4, 5). These levels induce total loss of equilibrium, stage II, within 31.0 to 0.8 minutes respectively, depending on temperature, species, and size of fish. Concentrations outside this range either were ineffective or produced narcosis at rates which would be relatively impractical for most fish-handling operations.

Methylpentynol is considerably less potent as a fish anesthetic than MS-222 (McFarland, 1959). MS-222 narcotizes the species used in this study within 2 to 3 minutes at concentrations of about 100 p.p.m. at 12° C. (Schoettger and Julin, 1967). We found that at least 5,000 p.p.m. of methylpentynol were required to produce a similar effect; this concentration anesthetized the most resistant individuals in

TABLE 2.--Efficacy of methylpentynol on two sizes of rainbow trout at three temperatures

Concentration (p.p.t.)	Temperature (°C.)	Fish		Mean induction time (minutes) for			Safety index		Recovery	
		Length (inches)	Number	All fish into		First fish into			Mean time (minutes)	Percent
				Stage A[1]	Stage B[2]	Stage C[3]	C/A[4]	C/B[5]		
1.5.....	17	2-6	20	31.0	81.5	46.0	1.5	0.6	8.0	65
1.5.....	17	7-12	20	18.0	19.0	15.5	0.9	0.8	6.3	95
2.0.....	12	2-6	10	23.0	52.5	35.8	1.6	0.7	17.0	90
Do.....	17	2-6	20	7.8	16.5	12.8	1.6	0.8	10.8	90
2.0.....	12	7-12	20	18.8	22.3	23.3	1.2	1.0	9.5	80
Do.....	17	7-12	20	6.3	9.0	8.3	1.3	0.9	5.0	100
3.0.....	12	2-6	10	8.3	15.8	15.8	1.9	1.0	17.0	100
Do.....	17	2-6	20	3.0	6.0	5.5	1.8	0.9	5.3	100
3.0.....	12	7-12	20	4.8	8.0	9.0	1.9	1.1	6.5	100
Do.....	17	7-12	20	3.3	5.0	4.5	1.4	0.9	6.0	100
4.0.....	7	2-6	20	12.5	19.8	20.3	1.6	1.0	27.8	100
Do.....	12	2-6	10	4.3	8.0	8.3	1.9	1.0	14.5	100
Do.....	17	2-6	20	2.0	2.8	2.8	1.4	1.0	5.0	100
4.0.....	7	7-12	20	11.5	18.8	14.0	1.2	0.7	15.3	100
Do.....	12	7-12	20	4.0	6.5	4.5	1.1	0.7	6.8	100
Do.....	17	7-12	20	1.8	3.0	3.5	2.0	1.2	6.8	100
5.0.....	7	2-6	20	8.5	12.3	12.0	1.4	1.0	32.8	100
Do.....	12	2-6	15	2.8	5.3	6.5	2.4	1.2	15.8	100
Do.....	17	2-6	20	1.3	2.3	2.5	2.0	1.0	8.5	95
5.0.....	7	7-12	20	8.3	9.8	9.0	1.1	0.9	13.8	100
Do.....	12	7-12	20	2.0	2.5	3.0	1.5	1.2	6.5	100
Do.....	17	7-12	20	1.3	2.5	2.8	2.2	1.1	6.8	100
6.0.....	7	2-6	20	4.3	7.3	8.5	2.0	1.2	25.5	100
Do.....	12	2-6	10	1.5	3.3	3.3	2.2	1.0	16.3	100
6.0.....	7	7-12	20	6.3	8.5	7.3	1.2	0.9	15.3	100
Do.....	12	7-12	20	1.5	2.0	2.5	1.7	1.3	7.0	100
7.0.....	7	2-6	20	4.3	7.8	6.8	1.6	0.9	28.3	100
Do.....	12	2-6	10	1.0	2.0	2.8	2.8	1.4	17.8	100
7.0.....	7	7-12	20	4.5	6.5	5.5	1.2	0.8	17.3	100
Do.....	12	7-12	20	1.3	1.8	2.5	2.0	1.4	8.3	100
8.0.....	7	2-6	20	3.3	4.5	5.5	1.7	1.2	25.8	90
Do.....	12	2-6	10	0.8	1.3	2.5	3.3	2.0	17.5	100
8.0.....	7	7-12	20	4.3	5.3	5.5	1.3	1.0	18.3	100
Do.....	12	7-12	20	1.0	1.8	2.3	2.2	1.3	8.5	100

[1] Total loss of equilibrium, stage II
[2] Total loss of reflex
[3] Medullary collapse
[4] Time for Stage C divided by time for Stage A
[5] Time for Stage C divided by time for Stage B

groups of 2- to 6-inch brown trout and lake trout within 3.5 minutes and the least resistant individuals among 7- to 12-inch rainbow trout and brown trout within 2 minutes. The greater sensitivity of the larger fish is even more apparent at lower concentrations (tables 2 and 3). In our opinion the size-sensitivity relation may not be valid. Large and small rainbow and brown trout were obtained from different sources (table 1); consequently interaction between size and strain may be reflected in the results.

TABLE 3.--Efficacy of methylpentynol on brown trout of two sizes at three temperatures

Concentration (p.p.t.)	Temperature (°C.)	Fish		Mean induction time (minutes) for			Safety index		Recovery	
		Length (inches)	Number	All fish into		First fish into			Mean time (minutes)	Percent
				Stage A[1]	Stage B[2]	Stage C[3]	C/A[4]	C/B[5]		
1.5.....	17	2-6	20	20.3	45.0	26.8	1.3	0.6	9.5	90
1.5.....	17	7-12	20	11.3	42.0	27.8	2.5	0.7	10.0	100
2.0.....	12	2-6	20	12.8	26.5	19.3	1.5	0.7	23.8	100
Do.....	17	2-6	20	6.8	15.5	12.8	1.9	0.8	6.5	95
2.0.....	12	7-12	20	11.8	51.0	28.3	2.4	0.6	38.0	85
Do.....	17	7-12	20	5.0	9.0	7.8	1.6	0.9	5.8	100
3.0.....	7	2-6	20	20.3	35.0	29.3	1.4	0.8	28.8	100
Do.....	12	2-6	20	10.0	13.8	11.3	1.1	0.8	17.3	90
Do.....	17	2-6	20	3.0	4.5	4.8	1.6	1.1	5.0	100
3.0.....	7	7-12	10	17.5	28.5	35.5	2.0	1.2	21.0	100
Do.....	12	7-12	20	6.0	9.5	9.3	1.5	1.0	11.0	100
Do.....	17	7-12	20	2.3	4.3	5.0	2.2	1.2	5.8	100
4.0.....	7	2-6	20	9.0	14.8	11.8	1.3	0.8	30.8	100
Do.....	12	2-6	20	5.0	7.0	5.5	1.1	0.8	12.8	100
Do.....	17	2-6	20	2.0	2.5	3.0	1.5	1.2	5.8	100
4.0.....	7	7-12	20	8.5	16.8	13.0	1.5	0.8	16.3	100
Do.....	12	7-12	20	4.0	8.5	7.0	1.8	0.8	10.3	100
Do.....	17	7-12	20	1.0	1.3	2.0	2.0	1.6	6.8	100
5.0.....	7	2-6	20	7.3	10.5	10.8	1.5	1.0	18.3	100
Do.....	12	2-6	20	3.5	4.8	5.0	1.4	1.1	12.8	100
Do.....	17	2-6	20	1.5	2.0	2.5	1.7	1.3	6.5	100
5.0.....	7	7-12	20	4.5	9.5	8.3	1.8	0.9	13.0	100
Do.....	12	7-12	20	2.0	3.8	4.5	2.3	1.2	11.8	100
Do.....	17	7-12	20	0.5	0.8	11.3	2.5	1.7	6.8	100
6.0.....	7	2-6	20	4.0	6.5	5.3	1.3	0.8	16.5	100
Do.....	12	2-6	20	1.8	2.5	2.5	1.4	1.0	8.5	100
6.0.....	7	7-12	20	3.0	7.5	5.8	1.9	0.8	11.0	100
Do.....	12	7-12	20	1.5	2.3	3.0	2.0	1.3	7.8	100
7.0.....	7	2-6	20	3.5	6.3	5.8	1.6	0.9	16.5	100
7.0.....	7	7-12	20	2.3	5.8	5.0	2.2	0.9	14.3	100
8.0.....	12	2-6	20	1.3	1.8	2.0	1.6	1.1	9.3	100
8.0.....	12	7-12	20	1.3	2.0	2.8	2.2	1.4	10.8	100

[1] Total loss of equilibrium, stage II
[2] Total loss of reflex
[3] Medullary collapse
[4] Time for Stage C divided by time for Stage A
[5] Time for Stage C divided by time for Stage B

Temperature had a pronounced effect on the efficacy of methylpentynol. To illustrate, a concentration of 5 p.p.t. produced loss of equilibrium in all species within 4.5 to 8.5 minutes at 7° C., within 2 to 3.5 minutes at 12°, and within 0.5 to 1.5 minutes at 17°. In general, a change in temperature of 5°C. changed anesthetizing time by a factor of approximately 2 to 3.

The tractability was not consistent among the four salmonids when anesthetized to total loss of equilibrium, stage II. Lake trout and large rainbow trout were relatively immobile. The other species and sizes of fish retained considerable reflex activity, and were difficult to handle during simulated stripping procedures. Also, a state of total loss of equilibrium was inadequate for delicate surgical

TABLE 4.--Efficacy of methylpentynol on brook trout at three temperatures

Concentration (p.p.t.)	Temperature (°C.)	Fish		Mean induction time (minutes) for			Safety index		Recovery	
		Length (inches)	Number	All fish into		First fish into			Mean time (minutes)	Percent
				Stage A[1]	Stage B[2]	Stage C[3]	C/A[4]	C/B[5]		
2.0.....	12	2-6	20	25.0	54.0	42.0	1.7	0.8	26.0	80
2.0.....	17	2-6	20	12.8	31.0	28.5	2.2	0.9	12.5	90
3.0.....	7	2-6	20	22.3	34.5	34.5	1.5	1.0	26.3	100
Do.....	12	2-6	20	13.3	18.8	16.0	1.2	0.9	23.3	100
Do.....	17	2-6	20	4.8	9.5	9.8	2.0	1.0	9.0	100
4.0.....	7	2-6	20	6.5	12.3	15.5	2.4	1.3	23.0	100
Do.....	12	2-6	20	3.5	10.8	10.0	2.9	0.9	19.0	100
Do.....	17	2-6	20	2.5	4.0	4.3	1.7	1.0	10.5	100
5.0.....	7	2-6	20	4.8	13.8	13.3	2.8	1.0	23.5	100
Do.....	12	2-6	20	2.5	7.3	4.3	1.7	0.6	12.5	100
Do.....	17	2-6	20	1.3	4.5	4.0	3.2	0.9	11.3	100
6.0.....	7	2-6	20	3.3	8.0	8.3	2.5	1.0	21.0	100
Do.....	12	2-6	20	1.8	6.8	4.5	2.6	0.7	15.8	100
8.0.....	7	2-6	20	2.5	5.5	4.8	1.9	0.9	17.3	95
Do.....	12	2-6	20	1.3	3.5	2.3	1.8	0.6	12.0	100

[1] Total loss of equilibrium, stage II
[2] Total loss of reflex
[3] Medullary collapse
[4] Time for Stage C divided by time for Stage A
[5] Time for Stage C divided by time for Stage B

TABLE 5.--Efficacy of methylpentynol on lake trout at three temperatures

Concentration (p.p.t.)	Temperature (°C.)	Fish		Mean induction time (minutes) for			Safety index		Recovery	
		Length (inches)	Number	All fish into		First fish into			Mean time (minutes)	Percent
				Stage A[1]	Stage B[2]	Stage C[3]	C/A[4]	C/B[5]		
1.5.....	17	2-6	20	31.5	69.8	30.8	1.0	0.4	11.0	60
2.0.....	7	2-6	10	33.5	84.5	41.0	1.2	0.5	57.3	100
Do.....	12	2-6	20	25.8	53.0	32.5	1.3	0.6	15.3	100
Do.....	17	2-6	20	9.5	23.8	10.8	1.1	0.5	7.5	50
3.0.....	7	2-6	20	12.8	25.3	19.8	1.5	0.8	26.8	100
Do.....	12	2-6	20	8.0	15.3	12.3	1.5	0.8	12.8	100
Do.....	17	2-6	20	1.8	3.3	3.0	1.7	0.9	7.5	35
4.0.....	7	2-6	20	9.0	13.5	12.3	1.4	0.9	17.5	100
Do.....	12	2-6	20	4.5	7.3	6.8	1.5	0.9	11.8	100
Do.....	17	2-6	20	0.8	0.8	1.3	1.7	1.7	4.3	80
5.0.....	7	2-6	20	5.8	5.8	8.0	1.4	1.4	14.3	100
Do.....	12	2-6	20	3.5	4.0	5.0	1.4	1.3	10.3	100
Do.....	17	2-6	20	0.5	0.5	1.0	2.0	2.0	7.0	85
6.0.....	7	2-6	20	3.0	3.0	5.3	1.8	1.8	12.3	100
Do.....	12	2-6	20	2.5	2.5	4.0	1.6	1.6	12.5	100
8.0.....	7	2-6	20	2.0	2.0	3.5	1.8	1.8	17.0	100
Do.....	12	2-6	20	1.0	1.0	1.5	1.5	1.5	12.3	100

[1] Total loss of equilibrium, stage II
[2] Total loss of reflex
[3] Medullary collapse
[4] Time for Stage C divided by time for Stage A
[5] Time for Stage C divided by time for Stage B

operations on rainbow trout. (Personal communication from Dr. Joseph B. Hunn, Fishery Biologist, Fish Control Laboratory, La Crosse, Wis., October 14, 1966.)

Complete immobility of the trout can be obtained by continuing exposure to the loss-of-reflex stage. With a concentration of 5 p.p.t. at 12° C., loss of reflex usually occurs less than 3 minutes after loss of equilibrium (tables 2, 3, 4, 5). It occurs more rapidly at 17° C. and more slowly at 7°. However, we must point out that the fish enter medullary collapse shortly after loss of reflex, particularly at higher concentrations and temperatures.

The relative risk of overexposing trout when attempting to maintain anesthesia is expressed by the safe exposure indexes shown in tables 2, 3, 4, and 5. The danger is obviously greater after the fish have entered loss of reflex, since the values are approximately 1.0 or less. In other words, in a given lot of fish, the most susceptible individuals are entering medullary collapse at the time the most resistant fish are entering loss of reflex. The exposure indexes for methylpentynol-treated trout do not appear to be correlated with concentration, temperature, species, or size of fish. By way of comparison, Schoettger and Julin (1967) reported safe exposure indexes of 2 to 3 for similar species and sizes of trout anesthetized in MS-222.

The mean times for recovery of fish from anesthesia ranged from 4 to 57 minutes (tables 2, 3, 4, 5). In general, recovery was most rapid at 17° C. and least rapid at 7°. Most mortalities among small rainbow trout and lake trout occurred after long exposures to low concentrations at 17°. In one instance, 35 percent of the rainbow trout died after a prolonged exposure to 1.5 p.p.t. The mortality of lake trout varied from 15 to 65 percent at this temperature, but the greatest mortalities occurred following exposures to 3 p.p.t., or less.

In all of the tests, we attempted to transfer the fish to well-oxygenated, circulating water for recovery just before their opercular movements ceased. With protracted exposures, the progression of deepening sopor in the more sensitive fish appears to become irreversible prior to medullary collapse. This was revealed in preliminary trials at 17° C. when deaths occurred among fish which were transferred to oxygen-rich water for recovery, even though they displayed opercular rhythm at the time of removal from the anesthetic. The mortality was even greater upon transfer to unaerated, static water.

In order to avoid the rapid onset of medullary collapse associated with high concentrations, it is necessary to reduce concentration, thus sacrificing rapid anesthesia for longer handling time.

As mentioned above, there also is a risk when exposing trout to low concentrations for long periods. However, the slower rates of anesthesia at these levels permit greater control over exposure than at higher concentrations.

A good compromise between the rate of anesthesia and handling time is achieved at concentrations between 2 and 4 p.p.t. at 12° C. These relations are demonstrated in figure 1. The greatest changes in time to loss of equilibrium, stage II, loss of reflex and medullary collapse with concentration occur between 2 and 4 p.p.t. Therefore, depending on the needs,

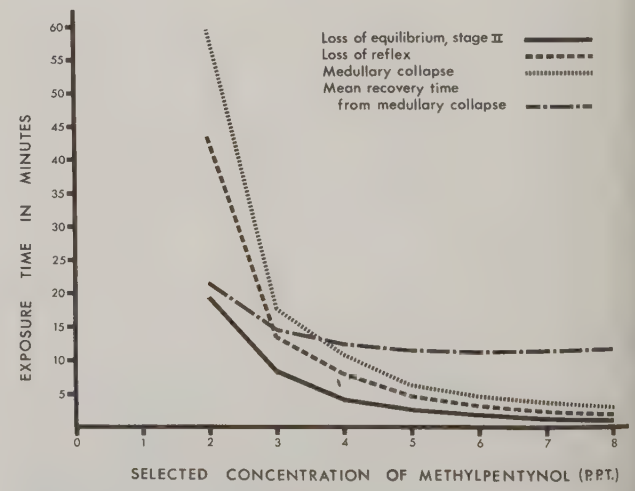

Figure 1.--Mean exposures required to induce various stages of anesthesia in trout at 12° C. for selected concentrations of methylpentynol. Curves for the response times were derived from the average of response times at each concentration for various species and sizes of trout.

the fishery worker can modify the concentration to produce the type or rate of anesthesia and handling time to meet his requirements. However, he also must consider the effects of temperature on efficacy in making an estimate of effective concentrations--a factor of about 2 to 3 occurs for each 5° C. change in temperature.

Effects of Water Quality

The efficacy of methylpentynol was neither enhanced nor decreased in bioassays on rainbow trout at 12° C. in solutions with pH's of 5, 7, and 9, and those with total hardnesses of 10, 42, and 180 p.p.m. as $CaCO_3$.

Repeated Use of Solutions

Five liters of a 5-p.p.t. solution of methylpentynol effectively anesthetized 260 rainbow trout, 7 to 12 inches long, at 12° C. The time required to anesthetize additional fish increased with each introduction. Thus, approximately 1 kilogram of rainbow trout can be anesthetized effectively per milliliter of drug. Meister and Ritzi (1958) reported that about 14 kilograms of brook trout and 42 kilograms of lake trout could be anesthetized per gram of MS-222.

Effects of Repeated Anesthetization

The repeated exposure of fish to methylpentynol had little influence on rate of response to the drug. From the third exposure on, the fish reacted violently upon contact with the solution, by thrashing and leaping before narcosis. Gross internal pathological examination disclosed no detrimental effects due to the anesthesia.

DISCUSSION

Our investigation establishes that methylpentynol is enigmatic as a fish anesthetic. It is capable of inducing profound stupor in four salmonids and yet it also appears incapable of producing consistently good anesthetization. The condition evoked by this drug seems closer to inebriation than anesthesia, as evidenced by such side effects as preanesthesia and postanesthesia lunging and untoward reactions upon reanesthetization.

We stress that intractability to handling at loss of equilibrium, stage II, is pronounced in some species. Large brown trout are difficult to grasp because of tail thrashing, and small rainbow trout, brown trout, and brook trout are affected to a lesser degree. Such behavior undoubtedly impedes a practical fish-handling routine.

We corroborate the findings of Bell (1964) and Klontz (1964) regarding the struggling of fish entering narcosis, incomplete immobility, and tendency toward respiratory arrest. An explanation for these reactions is suggested in the literature. When humans are given daily doses of methylpentynol in excess of 2 grams, the drug tends to accumulate, and symptoms of intoxication appear by the third or fourth day (Goodman and Gilman, 1965). Kennedy and Marley (1959) reported abnormal brain-wave activity with toxic phenomena during a 5-day regime in humans. Also, they found a correlation between the degree of electroencephalic abnormality and the amount of physical disturbance produced by the drug in patients with toxic manifestations. A high rise of acetylcholine in rat brains, related to the degree of depression, was reported by Pepeu (1960). Margolin et al. (1951) found that methylpentynol does not possess analgesic or anesthetic properties when tested on mice and dogs, and that it lacks antispasmodic action; it was proffered solely as a soporific in human medicine.

The nature and size of the operation should determine whether this relatively expensive drug is a practical anesthetic for use in fishery management. A preferred stage of anesthesia cannot be recommended because of the aforementioned shortcomings at each level.

CONCLUSIONS

1. Methylpentynol is effective for the anesthesia of rainbow trout, brown trout, brook trout, and lake trout at concentrations of 1.5 to 8 p.p.t.

2. The rate of anesthesia increases as the concentration and/or the water temperature increases. A 5 °C. change in water temperature causes a change in the rate of anesthesia by a factor of 2 to 3.

3. Concentrations from 2 to 4 p.p.t. appear better suited for handling of fish than do higher levels.

4. Water quality and pH of the anesthetic solution have no effect on the rate of anesthesia.

5. Methylpentynol does not produce the desirable or consistent characteristics of anesthesia which are acceptable for handling fish. Small lake trout and large rainbow trout were amenable to simulated stripping procedures, while small rainbow trout, small brook trout, and large and small brown trout were not.

6. Methylpentynol is less potent than MS-222 as a fish anesthetic. Fifty times as strong a solution of methylpentynol is necessary to yield the effect of a solution containing 100 p.p.m., of MS-222.

7. Methylpentynol appears to be more appropriate as a sedative or soporific for salmonids than as an anesthetic.

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INVESTIGATIONS IN FISH CONTROL

30. Toxicity of Methylpentynol to Selected Fishes

By Leif L. Marking

UNITED STATES DEPARTMENT OF THE INTERIOR
Fish and Wildlife Service
Bureau of Sport Fisheries and Wildlife
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CONTENTS

	Page
Abstract	3
Methods and materials	3
Results	4
Species and sizes of fish	4
Effects of temperature	6
Effects of water quality	6
Discussion	6
Conclusions	6
References	7

TOXICITY OF METHYLPENTYNOL TO SELECTED FISHES

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Abstract.--Methylpentynol was tested in 96-hour bioassays for its toxicity to rainbow trout, brown trout, brook trout, lake trout, northern pike, channel catfish, bluegills, largemouth bass, and walleyes. The LC50's range from 660 to 1,890 parts per million at 12° C. Channel catfish are the most resistant and lake trout are the most sensitive. Two-inch rainbow trout, brown trout, and lake trout are more sensitive to methylpentynol than larger ones in 96-hour exposures. The drug is more toxic to bluegills at 17° than at 12° C. Toxicity was not influenced in different water hardnesses of 10 to 180 parts per million.

Methylpentynol (3-methyl-1-pentyn-3-ol) has been used in human medicine as a hypnotic and sedative at oral doses of 100 to 800 milligrams (Hirsh and Orsinger, 1952) and 250 to 500 milligrams (Stecher et al., 1960). Overdoses, however, may produce acute psychosis, abnormalities of the nervous system, or coma and death in extreme cases. Since methylpentynol possesses no unique advantages as a human hypnotic to outweigh its disadvantages, it is no longer recommended (Sharpless, 1965). The oral LD50 for mice, rats, and guinea pigs is 600 to 900 mg./kg. (Margolin, Perlman, and McGavack, 1951).

Methylpentynol is a recognized anesthetic for fish at concentrations of 0.5 to 0.9 ml./1. (Klontz, 1964). Others have reported applications of the drug as a hypnotic in transporting fish at concentrations of 1 to 3 ml./gal. (Bell, 1964; Carlson, 1965; Fry and Norris, 1962; and McFarland, 1959 and 1960). Bayliff and Klima (1962) reported methylpentynol toxic to some fish at 6 ml./gal. (approximately 1,580 p.p.m.) and suggested it was too slow-acting for field use. Howland and Schoettger (1969) found that 1,500 to 8,000 p.p.m. of methylpentynol produced anesthesia within 4 to 57 minutes, but suggest the compound is better suited as a sedative than an anesthetic. As an anesthetic there is danger of overexposing the fish because of the high concentrations required to achieve rapid anesthesia.

Since fish can be overdosed with methylpentynol, we should determine its toxicity to fish. Amendments to the Federal Food, Drug, and Cosmetic act also require toxicity data to clear the drug for this use (Lennon, 1967). I have chosen to determine the toxicity of methylpentynol to rainbow trout, brown trout, brook trout, lake trout, northern pike, channel catfish, bluegills, largemouth bass, and walleyes with considerations to changes in temperature, water quality, and size of fish.

METHODS AND MATERIALS

Experimental fish were obtained from State and Federal fish hatcheries (table 1). Three size groups included 1- to 3-inch, 3- to 5-inch, and 6- to 9-inch fish. Ten fish were tested at each of 10 or 11 concentrations of methylpentynol in 15-liter, static bioassays according to the methods of Lennon and Walker (1964).

TABLE 1.--Fishes used in tests of methylpentynol

Common and scientific name	Source
Rainbow trout (Salmo gairdneri)	Manchester NFH[1], Iowa
Brown trout (Salmo trutta)	Manchester NFH, Iowa
Brook trout (Salvelinus fontinalis)	Osceola SFH[2], Wis.
Lake trout (Salvelinus namaycush)	Jordan River NFH, Mich. and St. Croix Falls SFH, Wis.
Northern pike (Esox lucius)	Garrison Dam NFH, N.D. and Gavins Point NFH, S.D.
Channel catfish (Ictalurus punctatus)	Fairport NFH, Iowa
Bluegill (Lepomis macrochirus)	Lake Mills NFH, Wis.
Largemouth bass (Micropterus salmoides)	Genoa NFH, Wis.
Walleye (Stizostedion vitreum)	Garrison Dam NFH, N.D.

[1] National Fish Hatchery
[2] State Fish Hatchery

At least 10 of the 1- to 3-inch fish served as controls. The bioassays with the 3- to 5-inch and 6- to 9-inch fish were conducted in polyethylene tanks containing 45 liters of aerated solution. Five concentrations were tested against the 9-inch fish and 10 fish served as controls.

Waters of different hardnesses and pH were prepared by adding greater or less amounts of reconstituting salts to deionized water (table 2). Temperatures of 7°, 12°, 17°, and 22° C. were maintained by placing the bioassay vessels in thermostatically controlled water baths. All temperatures are reported in Celsius.

Aliquots of Eastman-grade methylpentynol, purchased from Eastman Organic Chemicals, were pipetted into the bioassays to yield the desired concentrations.

The data on survival and mortality were recorded at 24, 48, and 96 hours and were analyzed according to the methods of Litchfield and Wilcoxon (1949) to determine LC50's, variations, slope functions, and 95-percent confidence intervals.

RESULTS

Species and Sizes of Fish

The LC50's of methylpentynol range from 660 to 1,890 p.p.m. in 96-hour bioassays for all species tested (table 3). Channel catfish, 2 and 4 inches long, are the most resistant species while small lake trout are the most sensitive.

Larger brown trout and lake trout are more resistant to the lethal effects of methylpentynol than the 2-inch ones. This is apparent with brown trout and lake trout at all observation periods, and with one lot of rainbow trout at 96 hours. The medium sized brown trout and lake trout tolerate concentrations which approximate those tolerated by the larger individuals. LC50's range from 1,060 to 1,160 p.p.m. in 96 hours for 3- to 7-inch brown trout and lake trout.

Three- and 4-inch brook trout responded uniformly to methylpentynol with 96-hour LC50's of 1,200 and 1,280 p.p.m. respectively. However, the 6-inch fish appear more sensitive in the 96-hour bioassay. This may be explained by the fact that they contracted furunculosis just prior to the bioassays. Unfortunately, the disease was not diagnosed until after the tests.

Methylpentynol acted slowly at the concentrations tested, and little or no mortality occurred within 3 hours, even at concentrations

TABLE 2.--Water qualities obtained with different amounts of reconstituting salts in deionized water

Classification of water	Salt added in mg./l.				pH range	Concentration as p.p.m. $CaCO_3$	
	$NaHCO_3$	$CaSO_4$	$MgSO_4$	KCl		Total hardness	Total alkalinity
Soft	12	7.5	7.5	0.5	6.4-6.8	10-13	10-13
Standard[1]	48	30.0	30.0	2.0	7.2-7.6	40-48	30-35
Medium	192	120.0	120.0	8.0	7.6-8.0	160-180	110-120

[1] Standard reconstituted water used in routine bioassay.

TABLE 3.--Toxicity of methylpentynol to nine species of fish at 12° C.

Species	Average weight (grams)	Approximate length (inches)	LC50 (p.p.m.) and 95-percent confidence interval at		
			24 hours	48 hours	96 hours
Rainbow trout	1.9	2	1,220 1,151-1,294	1,150 1,095-1,208	870 753-1,005
Do	1.6	2	1,340 1,276-1,407	1,280 1,219-1,344	1,250 1,214-1,288
Do	23.0	6	1,300 1,204-1,404	1,290 1,217-1,367	1,260 1,223-1,298
Brown trout	2.6	2	820 745-902	750 714-786	680 636-728
Do	14.3	4	1,090 1,048-1,134	1,085 1,033-1,139	1,060 1,010-1,113
Do	27.0	6	1,130 1,100-1,158	1,100 1,028-1,177	1,100 1,038-1,166
Brook trout	12.5	3	1,300 1,250-1,352	1,270 1,221-1,321	1,200 1,132-1,272
Do	20.0	4	1,375 1,317-1,430	1,300 1,250-1,352	1,280 1,231-1,331
Do	37.5	6	1,210 1,142-1,283	1,175 1,108-1,246	1,100 1,028-1,177
Lake trout	2.0	2	900 849-954	860 789-937	660 584-746
Do	5.6	3	1,280 1,143-1,434	1,230 1,108-1,365	1,160 1,084-1,241
Do	35.0	7	1,220 1,140-1,305	1,200 1,132-1,272	1,160 1,094-1,230
Northern pike	1.8	2	1,050 1,012-1,077	1,000 943-1,060	<900 ---
Channel catfish	1.9	2	1,770 1,702-1,841	1,770 1,702-1,841	1,700 1,604-1,802
Do	5.2	4	1,890 1,817-1,966	1,890 1,817-1,966	1,890 1,817-1,966
Bluegill	1.3	2	1,390 1,337-1,446	1,340 1,301-1,380	1,340 1,301-1,380
Do	2.8	3	1,370 1,317-1,425	1,350 1,320-1,382	1,260 1,189-1,336
Largemouth bass	0.5	1	1,250 1,220-1,282	1,250 1,202-1,282	1,170 1,114-1,228
Do	5.2	4	1,270 1,233-1,308	1,185 1,129-1,244	1,100 1,028-1,177
Do	63.0	7	1,400 1,321-1,484	1,300 1,250-1,352	1,250 1,190-1,312
Walleye	0.7	2	1,225 1,156-1,298	1,160 1,048-1,288	1,140 1,027-1,265

substantially higher than those required to kill fish at 24 hours. Little additional mortality occurred among any species after 24 hours, except the small rainbow trout, brown trout, and lake trout. In fact, the LC50's are identical, or nearly identical at 24, 48, and 96 hours for some species.

Northern pike appear the most sensitive among the warmwater species. The young pike require a large supply of food and become predacious in the bioassay since no food is available. Also they become weak when not fed for 96 hours and a complete statistical evaluation could not be calculated at that time.

TABLE 4.--Toxicity of methylpentynol to rainbow trout and bluegills at different temperatures

Species	Temperature (°C.)	LC50 (p.p.m.) and 95-percent confidence interval at		
		24 hours	48 hours	96 hours
Rainbow trout.....	7	1,450 1,343-1,566	1,330 1,255-1,410	1,330 1,255-1,410
Do.....	12	1,340 1,276-1,407	1,280 1,219-1,344	1,250 1,214-1,288
Do.....	17	--	--	930 845-1,023
Bluegills.....	12	1,390 1,337-1,446	1,340 1,301-1,380	1,340 1,301-1,380
Do.....	17	1,120 1,040-1,198	1,070 1,009-1,334	1,030 928-1,143

Effects of Temperature

Methylpentynol is more toxic to 1.6-inch rainbow trout and 1.3-inch bluegills at the higher temperatures in 24- to 96-hour bioassays (table 4). The 96-hour LC50 for rainbow trout at 7° C. is 1,330 p.p.m. whereas at 17° C. the value drops to 930 p.p.m. Although bluegills are less sensitive, the toxicity of the drug increases about 20 percent with an increase in temperatures from 12° to 17° C. The LC50's for bluegills are 1,340 and 1,030 p.p.m. at 12° and 17° C. respectively at 96 hours. Little additional mortality of either species occurred after 24 hours and LC50's are fairly consistent throughout the bioassay.

Effects of Water Quality

Methylpentynol appears equally toxic to 1.9-inch rainbow trout in water containing about 11, 44, and 170 p.p.m. of total hardness at 24 and 48 hours and appears only slightly more toxic in the harder water at 96 hours (table 5). The difference appears insignificant in soft and standard water hardness at all observation periods. The LC50's ranged from 780 to 920 in the three water hardnesses at 96 hours.

TABLE 5.--Toxicity of methylpentynol to rainbow trout in selected water qualities[1] at 12° C.

Water hardness[1]	LC50(p.p.m.) and 95-percent confidence interval at		
	24 hours	48 hours	96 hours
Soft.....	1,260 1,202-1,320	1,200 1,135-1,268	920 821-1,018
Standard.	1,230 1,157-1,308	1,190 1,127-1,256	980 905-1,060
Medium...	1,210 1,163-1,258	1,200 1,138-1,265	780 603-1,009

[1] Water quality described in table 2.

DISCUSSION

Rainbow trout and bluegills are more sensitive to the toxic effects of methylpentynol at higher temperatures. This corresponds with efficacy trials of Howland and Schoettger (1969) in which higher temperatures stimulate the metabolic activity and the drug is assimilated much faster.

Anesthesia is induced by 1,500 to 8,000 p.p.m. of methylpentynol (Howland and Schoettger, 1969). These concentrations exceed LC50's for 24- to 96-hour bioassays, but the exposure time is only 4 to 57 minutes for effective anesthesia.

Parkhurst and Smith (1957) exposed rainbow trout to 2,400 p.p.m. of methylpentynol. Undoubtedly this concentration would become toxic to the trout in longer exposures.

CONCLUSIONS

The 96-hour LC50's of methylpentynol to fish range from 660 to 1,890 p.p.m. Higher concentrations are necessary to produce effective anesthesia, but exposures are also much shorter.

Larger brown trout and lake trout are more resistant to the toxic effects of methylpentynol than smaller ones.

The toxic effects of methylpentynol are manifest in 24 hours and little additional mortality occurs after this time.

Channel catfish are the most resistant and lake trout the most sensitive to methylpentynol.

Methylpentynol is more toxic to rainbow trout and bluegills at higher temperatures.

Changes in water hardnesses of approximately 12, 44, and 170 p.p.m. influence the toxicity of methylpentynol very little.

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INVESTIGATIONS IN FISH CONTROL

31. Annotated Bibliography on Methylpentynol

By Gerald E. Svendsen

UNITED STATES DEPARTMENT OF THE INTERIOR
Fish and Wildlife Service
Bureau of Sport Fisheries and Wildlife
Washington, D.C. · February 1969

ANNOTATED BIBLIOGRAPHY ON METHYLPENTYNOL

By Gerald E. Svendsen, Biologist
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Abstract.--An annotated bibliography containing 26 selected references on the biochemistry, physiology, and methods of analysis of methylpentynol.

Experiments with methylpentynol as an anesthetic for four salmonids to describe its toxicity and efficacy began in 1964 at the Fish Control Laboratory, La Crosse, Wis. The U.S. Food and Drug Administration requires these data for clearance and labeling of drugs. During the study, a number of selected references on fishery uses of the drug, and on its biochemistry, physiology, and methods of analysis were annotated.

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Bayliff, William H., and Edward F. Klima.
1962. Live-box experiments with anchovetas, Cetengraulis mysticetus, in the Gulf of Panama. Inter-American Tropical Tuna Commission Bulletin, vol. 6, No. 8, p. 333-436.

Four anesthetics--quinaldine, MS-222, methylpentynol, and tertiary amyl alcohol--were used to facilitate tagging operations. Quinaldine caused some mortalities and proved to be chemically unstable. Methylpentynol and tertiary amyl alcohol also killed some fish and acted too slowly for field use. The methylpentynol was used at a concentration of 6 milliliters per gallon. MS-222 is the most satisfactory of the chemicals tested, although there were some mortalities at the higher concentrations.

Bell, Gordon R.
1964. A guide to the properties, characteristics, and uses of some general anaesthetics for fish. Fisheries Research Board of Canada, Bulletin 148. 4 p.

The structural formulas, manufacturers, dosages, toxicities, and solubilities are presented for 11 chemicals used as fish anesthetics: carbon dioxide, chloral hydrate, chloretone, ether, methylpentynol, MS-222, phenoxyethanol, quinaldine, sodium amytol, tribromomethanol, and tertiary amyl alcohol. Methylpentynol is recommended at a dosage of 1 to 2 milliliters per liter in transportation. Bell does not recommend it as an anesthetic for surgery because immobilized fish twitch when prodded or cut. Methylpentynol immobilizes fish slowly with occasional violent struggling, but recovery is rapid in fresh water.

Carlson, Frank T.
1965. Susquehanna River Shad study. Pennsylvania Angler, October, p. 1-7.

Shad were transported in a solution containing 1 milliliter of methylpentynol per gallon of water. They were anesthetized in a 15-gallon tub of river water containing 3 milliliters of methylpentynol per gallon.

Fry, F. E. J., and K. S. Norris.
1962. The transporting of live fish. In Georg Borgstrom, Editor Fish as Food, Vol. 2, Chap. 17, p. 595-608. Academic Press, N.Y.

The authors suggest methylpentynol for use in transporting live fish.

Hirsh, Harold L., and William H. Orsinger.
1952. Methylparafynol- a new hypnotic. Preliminary report on its therapeutic efficacy and toxicity. American Practioner, vol. 3, no. 1, p. 23-26.

Patients received a 100 to 800 milligram dose of methylpentynol as a sedative with no after effects on blood pressure, pulse, respiration rate, blood and urine composition, electrocardiogram, or liver and kidney function. The authors considered methylpentynol a safe, nontoxic, efficient, rapid, and long lasting hypnotic drug in humans.

Howland, Robert M., and Richard A. Schoettger.

1969. Investigations in Fish Control: 29. The efficacy of methylpentynol as an anesthetic on four salmonids. U.S. Bureau of Sport Fisheries and Wildlife.

Methylpentynol was tested as an anesthetic against rainbow trout, brown trout, brook trout, and lake trout. Concentrations ranging from 1.5 to 8.0 parts per thousand produced anesthesia within 4 to 57 minutes respectively. They studied the effects of water temperature, water quality, and pH on the rate of anesthetization and found that only temperature had a measurable effect. The authors concluded that methypentynol is better suited as a sedative than as an anesthetic for salmonids, because fish under anesthesia are not completely immobilized.

Job, C. von.

1959. Die Beziehungen zwischen der stärke der narkotischen wirkung und der thermodynamischen konzentration bei estern des methylpentinols. Arzneimittel-Forschung, vol. 9, no. 1, p. 14-22.

The author investigated the effects of methylpentynol and some of its esters on the rat and on the isolated frog nerve. Action potentials of the isolated frog nerve are retarded and interrupted under the influence of methylpentynol. Methylpentynol blocks conduction of stimuli at a concentration of 5 grams per liter.

Kennedy, Walter A., and Edward Marley.

1959. The electroencephalographic effects of methylpentynol. Electroencephalography and Clinical Neurophysiology, vol. 2, no. 1, p. 59-64.

The authors discuss several aspects of the drug in clinical work, and cite cases of overdosage and side effects from methylpentynol. They found a correlation between the degree of electroencephalographic abnormalities and the amount of physical disturbance produced in patients given 0.5 gram of methylpentynol orally for 5 days.

Klontz, George W.

1964. Anesthesia of fishes. In Proceedings of the Symposium on Experimental Animal Anesthesiology. Brooks Air Force Base, December 14-16. 13 p.

Techniques for anesthetizing fish and a brief description are given for 15 agents: carbon dioxide, electricity, diethyl ether, secobarbital sodium, amobarbitol sodium, urethane, chloral hydrate, tertiaryamyl alcohol, tribromoethanol, chlorobutanol, 2-phenoxyethanol, 4-styrylpyridine, methylpentynol, quinaldine, and MS-222. Concentrations of 0.5 to 0.9 milliliter per liter of methylpentynol are suggested to induce anesthesia within 2 to 3 minutes. Maintenance of anesthesia is considered fair, and the recovery of fish in fresh water occurs in 5 to 20 minutes. The author also rated the maintenance of deep anesthesia as fair because fish seem to go into respiratory arrest. He considered the odor of methylpentynol quite disagreeable.

Lasagna, Louis.

1954. A comparison of hypnotic agents. The Journal of Pharmacology and Experimental Therapeutics, vol. III, p. 9-20.

The author used chloral hydrate, pentobarbital sodium, methylpentynol, and a placebo to determine which doses are most useful for inducing prolonged sleep in man. A dosage of 0.5 to 1.0 gram of methylpentynol induced sleep that was undistinguishable from that of the placebo.

Leal, Aluiso Marques, and Maria Helena Diniz.

1956. Assay of methylpentynol in galenic preparations. Revista Portuguesa de Farmacia, vol. 6, p. 14-17.

Two titrimetric methods for the assay of methylpentynol in body fluids are described.

Margolin, S., P. Perlman, F. Villani, and T. H. McGavack.

1951. A new class of hypnotics: unsaturated carbinols. Science, vol. 114, p. 384-385.

Methylpentynol was studied for use as a clinical hypnotic. The LC_{50} for mice, rats, and guinea pigs is 600 to 900 mg./kg. (milligrams per kilogram). The animals died in coma. Two hundred to 300 mg./kg. had no effect on mice, rats, and dogs. One half to 4.6 percent of a 200-mg./kg. dose fed to dogs was excreted in the urine within 24 hours. Ten minutes after an intravenous administration of 200 mg./kg. to dogs, 20 percent of the dose is found in the blood, but none is present after 2 hours. Tweny percent of an 800-mg./kg. dose is found in the muscle and liver tissues taken from rats which are still under hypnosis. No residues were found in these tissues after the effects of anesthesia wore off. Methylpentynol was not metabolized by whole blood of dogs or rats but it was metabolized by slices of kidney, liver, or brain.

Marking, Leif L.
1969. Investigations in Fish Control: 30. The toxicity of methylpentynol to selected fishes. U.S. Bureau of Sport Fisheries and Wildlife.

Toxicity of methylpentynol to rainbow trout, brown trout, brook trout, lake trout, northern pike, channel catfish, bluegills, largemouth bass, and walleyes of various sizes ranged from the 96-hour LC_{50} value of 660 p.p.m. (parts per million), for the more sensitive lake trout to 1,890 p.p.m. for channel catfish at 12° C. Larger rainbow, brook, and lake trout were considerably more resistant than smaller ones. Rainbow trout and bluegills are more sensitive to methylpentynol in warmer temperatures. Total hardnesses of 10.0 to 170.0 p.p.m. produced similar results in the static bioassays. Methylpentynol is much less toxic than other anesthetics tested in the 24-, 48-, and 96-hour static bioassays at selected temperatures and water qualities.

Marley, E.
1959. Pharmacology of methylpentynol and methylpentynol carbamate. British Journal of Pharmacology, vol. 14, p. 284-306.

Methylpentynol and methylpentynol carbamate are depressants of monosynaptic and polysynaptic reflexes in cats, frogs, rabbits, and guinea pigs. Small doses exerted weak ganglionic and neuromuscular blocking actions, increased aortic blood flow, diminished systolic amplitude, increased coronary flow, and stimulated respiration. Large doses depressed respiration.

Marley, E., and W. D. M. Paton.
1959. The effect of methylpentynol and methylpentynol carbamate on the perfused superior cervical ganglion of the cat. British Journal of Pharmacology, vol. 14, p. 303-312.

The output of acetylcholine in the perfused cervical ganglion is depressed by dosages of 1 to 5 milligrams of the drugs.

Marley, E., and J. R. Vane.
1958. The distribution of methylpentynol and methylpentynol carbamate in tissues and body fluids of cats. British Journal of Pharmacology, vol. 13, p. 364-371.

The authors present a modified titrimetric method for estimating methylpentynol in amounts as small as 0.1 milligram. They found no difference between plasma concentrations and whole blood concentrations of methylpentynol 10 minutes after injection. They concluded that the drug has free access to all parts of the body, and general anesthesia does not inhibit its metabolism and excretion. It enters cells easily, where it tends to accumulate.

Marley, Edward.
1958. Susceptibility to methylpentynol and methylpentynol carbamate. British Medical Journal, Medical Memoranda, August 23, p. 493.

He compared the toxicities of methylpentynol and methylpentynol carbamate in man. Methylpentynol at 0.5 gram per day for 5 days causes many toxic effects in man.

McFarland, William N.
1959. A study of the effects of anesthetics on the behavior and physiology of fishes. Publications of the Institute of Marine Science, University of Texas, vol. 6, p. 23-55.

The anesthetic effects of 21 chemicals on Fundulus parvipinnis, Gambusia affinis, Paralabrax alathratus, and Girella nigricans were investigated. The effects on behavioral patterns are observed in four major stages: sedation, loss of equilibrium, loss of reflex,

and medullary collapse. These stages are compared to the sequence of anesthesia described for higher vertebrates. Narcotic potencies are correlated with molecular weight of the drugs. Methylpentynol is rated highly potent.

McFarland, William N.
1960. The use of anesthetics for the handling and the transport of fishes. California Fish and Game, vol. 46, no. 4, p. 407-431.

The author suggested that MS-222, tertiary amyl alcohol and methylpentynol are beneficial for inducting deep anesthesia because the drugs act quickly and the fish recover rapidly. Recovery is complete, provided the respiratory movements have not ceased for more than a few minutes. A concentration of 1.5 to 2.0 milliliters per gallon of methylpentynol is considered desirable for transporting marine and freshwater fishes. Methylpentynol lowers metabolic rates and therefore increases load capacity. He suggests that fishes should be pretreated in the anesthetic to reduce metabolic rates prior to loading and transportation.

Nicholls, J. G., and J. P. Quilliam.
1956. The mechanism of action of paraldehyde and methylpentynol on neuromuscular transmission in the frog. British Journal of Pharmacology, vol. 11, p. 151-155.

Paraldehyde and methylpentynol block neuromuscular transmission by decreasing the secretion of acetyl cholinesterase at the synapse in the frog.

Norris, Kenneth S., Frank Brocato, Frank Calandrino and William N. McFarland.
1960. A survey of fish transportation methods and equipment. California Fish and Game, vol. 46, no. 1, p. 5-33.

Methylpentynol is suggested as a useful anesthetic in fish transportation.

Parkhurst, Z. E., and M. A. Smith.
1957. Various drugs as aids in spawning rainbow trout. The Progressive Fish-Culturist, vol. 19, no. 1, p. 39.

A methylpentynol concentration of 2,400 p.p.m. caused sluggishness and relaxation in rainbow trout. The trout are ready for spawning in 3.5 minutes at a water temperature of 43° F. Somewhat longer exposures are not harmful. Trout remained in good condition 75 days after spawning. There was an 84.1 percent hatch from those fish spawned with drugs, and an 84.0 percent hatch from controls.

Pepeu, Giancarlo, and Nicholas J. Giarman.
1960. Effect of methylpentynol on acetylcholine in the rats brain. Nature, vol. 186, p. 638.

The authors found that methylpentynol did not interfere with the synthesis of acetylcholine in the rat brain, nor did it inhibit cholinesterase activity. Male rats were given intraperitoneal injections of methylpentynol in a dosage varying from 200 to 500 milligrams/kilogram.

Perlman, Preston L., and Carol Johnson.
1952. The metabolism of Dormison (3-methyl-pentyne-ol-3, methyl-parafynol) and methods for the estimation of Dormison in biological materials. Journal of the American Pharmaceutical Association, vol. 41, no. 1, p. 13-16.

The authors described a tritrimetric method for estimating methylpentynol in biological fluids and tissue. They studied the metabolism of methylpentynol in dogs. Methylpentynol is metabolized by destruction of the ethinyl group, which metabolizes quite rapidly. This is revealed by the rapid decline in the blood level of methylpentynol and the slow level of elimination of the chemical in the urine, and in vitro by the disappearance of the ethinyl group by metabolizing rat tissues. They found no evidence of storage or accumulation of the drug in tissues.

Perlman, Preston L., David Sutter, and Carol B. Johnson.
1953. Further studies on the metabolic deposition of Dormison (3-methyl-1-pentyn-3-ol) in dogs and man. Journal of the American Pharmaceutical Association, vol. 42, no. 2, p. 750-753.

Dogs eliminated 17-27 percent of the administered methylpentynol conjugated with glucuronic acid; 1 percent unchanged in the urine, and none by way of the lungs or feces.

In man, up to 10 percent is eliminated unchanged, and 17-27 percent as conjugates of glucuronic acid. The peak levels for elimination of the drug are reached within 48 hours.

Quilliam, J. P.
1959. Paraldehyde and methylpentynol and ganglionic transmission. British Journal of Pharmacology, vol. 14, p. 277-283.

Paraldehyde and methylpentynol block transmission of the impulse at preganglionic nerve terminals in cats. The author suggests that this is caused by a decrease in the secretion of acetycholine from the preganglionic nerve terminals.

Schafferzich, S., and Beverly J. Brown.
1952. Anticonvulsant activity and toxicity of methylparafynol (Dormison) and some other alcohols. Science, vol. 116, p. 663.

The authors used phenobarbitol, 2-methyl-2-propanol, 2-methyl-2,4-pentamediol, 3-pentanol, 2-methyl-2-butanol, and methylpentynol as an anticonvulsant in treatment of epilepsy. Rats were not effected by 0.17 percent methylpentynol in their drinking water for 4 months. The LD_{50} of methylpentynol to rats is 300 to 900 milligrams per kilogram. They concluded that methylpentynol is the least safe of those chemicals studied.

Sheldon, J. M.
1965. Plastic bag transport of salmon and steelhead by air and car. The Progressive Fish-Culturist, vol. 27, no. 2, p. 86.

The author used methylpentynol, 0.67 milliliter per gallon of water, as a sedative to transport salmon and steelhead in plastic bags. Three milliliters of 10 percent Dow-Corning Antifoam emulsion was added to prevent excessive foaming.

Smith, J. N., and R. T. Williams.
1954. The metabolism of aliphatic alcohols. The glucuronic acid conjugation of chlorinated and some unsaturated alcohols. Biochemistry Journal, vol. 56, p. 618-621.

They studied the conjugation of glucuronic acid with a number of chlorinated and other aliphatic alcohols in the rabbit. About 50 percent of the methylpentynol is conjugated with glucuronic acid and excreted in the urine.

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19. Toxicity of Bayer 73 to Fish, by Leif L. Marking and James W. Hogan. 1967. 13 p.
20. Toxicity of Dimethyl Sulfoxide (DMSO) to Fish, by Wayne A. Willford. 1967. 8 p.
21. Labor-Saving Devices for Bioassy Laboratories, by Robert J. Hesselberg and Ralph M. Burress. 1967. 8 p.

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